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☐	L8	(EHEC or enterohemorrhagic) same vero	47
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☐	L6	coli same (vaccine or vaccines or vaccination or immunization or immunise or vaccinate) same (shed$ or coloniz$)	212
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☐	L4	L3 same coli	72
☐	L3	culture same supernatant same vaccin$	1281
☐	L2	(enterohemorrhagic or enterohaemorrhagic) same culture same supernatant	5
☐	L1	coli same vaccin$ same (enterohemorrhagic or enterohaemorrhagic)	54

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0013521797 BIOSIS NO.: 200200115308
Diarrhoeagenic Escherichia **coli**: An emerging problem?
AUTHOR: Clarke Stuart C (Reprint)
AUTHOR ADDRESS: Scottish Meningococcus and Pneumococcus Reference Laboratory, Department of Microbiology, North Glasgow University Hospital NHS Trust, Stobhill Hospital, Balornock Road, House on the Hill, Glasgow, G21 3UW, UK**UK
JOURNAL: Diagnostic Microbiology and Infectious Disease 41 (3): p93-98
November, 2001 **2001**
MEDIUM: print
ISSN: 0732-8893
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: Diarrhea remains one of the main sources of morbidity and morbidity in todays world and a large proportion is caused by diarrheagenic Escherichia ***coli*** . They are a particuar problem in developed countries although traveler's diarrhea and hemorrhagic colitis are also a problem in developed countries. There are seven classes of diarrheagenic E. ***coli*** , namely enteropathogenic E. ***coli*** (EPEC), ***enterohaemorrhagic*** E. ***coli*** (***EHEC***), enteroinvasive E. ***coli*** (EIEC), enterotoxigenic E. ***coli*** (ETEC), enteroaggregative E. ***coli*** (EAggEC), diarrhea-associated hemolytic E. ***coli*** (DHEC) and cytolethal distending toxin (CDT)-producing E. ***coli*** . Many of their virukence determinants have been determined and some classes of diarrheagenic E. ***coli*** produce toxins. The virulence factors of some diarrhogenic E. ***coli*** have yet to be full determined and in the meantime they remain a large and emerging problem without the availability of effective ***vaccines*** .

L7: Entry 4 of 9 File: PGPB Oct 31, 2002

DOCUMENT-IDENTIFIER: US 20020160020 A1
TITLE: Enterohemorrhagic escherichia coli vaccine

Brief Description of Drawings Paragraph:
[0047] FIG. 7 shows the number of animals shedding E. coli O157:H7 on each day of the vaccine trial described in Example 6. Bacteria were detected by direct plating of fecal samples which had been resuspended in saline on Sorbitol MaConkey agar supplemented with cefixime and tellurite. Solid bars, placebo group; hatched bars, EHEC vaccine group.

Brief Description of Drawings Paragraph:
[0049] FIG. 9 shows the percentage of each group of animals shedding E. coli O157:H7(Panel A) and the total number of bacteria recovered (Panel B) on each day of the trial described in Example 6. Bacteria were detected in feces by plating on Sorbitol MaConkey agar supplemented with cefixime and tellurite following immunomagnetic enrichment as described in J. Van Donkersgoed et al., Can. Vet. J. (2001) 42:714. (A) Solid bars, placebo; hatched bars, EHEC vaccine; open bars, .DELTA.Tir vaccine. (B) .box-solid., placebo group; .circle-solid., EHEC vaccine; .tangle-solidup., .DELTA.Tir vaccine.

Detail Description Paragraph:
[0131] Following oral challenge with E. coli O157:H7 on day 49, each group was monitored daily for fecal shedding of the organism for 14 days. In this experiment, bacteria were cultured following immunomagnetic enrichment (J. Van Donkersgoed et al., Can. Vet. J. (2001) 42:714, Chapman and Siddons, J. Med. Microbiol. (1996) 44:267) rather than direct plating since yearling cattle shed less than calves in this infection model. On the day of challenge, two animals in the placebo group were culture-positive for E. coli O157:H7 and were eliminated from the trial. The placebo-immunized animals shed the organism after challenge much more than those in the two EHEC vaccine groups (FIG. 9). Those which received the placebo vaccine shed the organism for a median of 4 days, significantly longer than the median of 0 days by the other two vaccine groups (p=0.0002, Kruskal-Wallis ANOVA). Significantly fewer bacteria were recovered from the EHEC and .DELTA.Tir vaccine groups (p=0.04, Kruskal-Wallis ANOVA). From day 2 post-infection onwards, 78% of the placebo animals shed the organism for at least one day as compared to 15% of the EHEC and 30% of the .DELTA.Tir vaccinates (Table 4).

Detail Description Paragraph:
[0133] Tir is likely required for colonization of the bovine intestine, and this is supported by the observation that a vaccine containing secreted proteins from a .DELTA.Tir E. coli O157:H7 strain was not as efficacious as an identical formulation from an isogenic wild-type isolate. However, the former vaccine was significantly more efficacious than a placebo suggesting that immunity against colonization is multifactorial in nature. This is supported by the Western blot analysis of the response to immunization in which several protein components as well as lipopolysaccharide were recognized. The contribution to protection by lipopolysaccharide is not known, but the presence of antibodies against this molecule does not correlate with protection in a murine EHEC model (Conlan et al., Can. J. Microbiol. (1999) 45:279; Conlan et al., Can. J. Microbiol. (2000) 46:283). Also, immunization with recombinant Tir and EspA can reduce numbers of bacteria

shed, but not the actual numbers of animals nor the duration of shedding.

Detail Description Table CWU:
4TABLE 4 Number of animals shedding E. coli O157:H7 at any time between day 2 and day 14 post-challenge. Number Percent Vaccine Shedding n Shedding p-value EHEC 2 13 15.4 0.003 .DELTA.Tir 3 10 30 0.008 Placebo 18 23 78.3 1

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L7: Entry 9 of 9 File: USPT Jun 22, 1999

DOCUMENT-IDENTIFIER: US 5914114 A
TITLE: Method of raising antibodies against E. coli of the family CS4-CFA/I

Brief Summary Text (4):
The effect of E. coli in mammals is dependent on the particular strain of organism. Many beneficial E. coli are present in the intestines. Since the initial association with diarrheal illness, five categories of diarrheagenic E. coli have been identified and are presently recognized: enterotoxigenic (ETEC), enteropathogenic (EPEC), enterohemorrhagic (EHEC), enteroaggregative (EAggEC), and enteroinvasive (EIEC). These categories are grouped according to characteristic virulence properties, such as elaboration of toxins and colonization factors and/or by specific types of interactions with intestinal epithelial cells. ETEC are the most common of the diarrheagenic E. coli and pose the greatest risk to travelers. E. coli of the family CS4-CFA/I are some of the more common enterotoxigenic E. coli. There is need for vaccines which are specific against this class of E. coli that give rise to antibodies that cross-react with and cross-protect against the more common members of the CS4-CFA/I family. There are six members of this family of ETEC fimbrial proteins, CFA/I, CS1, CS2, CS4, CS17 and PCF 0166. ETEC are responsible for high infant mortality in developing countries, with an estimate that almost 800,000 deaths per year due to these organisms. These organisms also cause illness in adult travelers to regions where the disease is endemic. No licensed vaccine exists against these organisms. The present vaccines being tested present problems related to manufacturing. So far, there has been no demonstration of significant efficacy of the prior art vaccines.

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L8: Entry 3 of 47 File: PGPB May 26, 2005

DOCUMENT-IDENTIFIER: US 20050112649 A1
TITLE: Sensitive and self-calibrating multi-test system the detection and identification of enterohemorrhagic escherichia coli (EHEC)

Summary of Invention Paragraph:
[0013] Besides the above-mentioned tests, in vitro tests are used to provide evidence of the expression of the main virulence factors in putative isolated EHEC strains (i.e., toxin and enterohemolysins). Detection of the toxins is carried out by the Vero cell assay.sup.(4). Immunological tests (i.e., immunoagglutination test kit or Enzyme-liked-immunosorbent-assay; ELISA) are also used to provide such evidence. On the other hand, the production of enterohemolysins is evidenced by the lysis of washed sheep red blood cells by in vitro testing. Though these tests are reliable when positive results are obtained, their interpretation is uncertain when they yield negative results since the strain may carry the corresponding genes without being able to express them under certain conditions. Therefore, further confirmation by genetic characterization is usually needed and results in additional labor, cost and time.

Detail Description Paragraph:
[0027] The technique will use the specific binding of shiga toxins to the Gb3 receptor as means to detect the toxins. These toxins have been detected either directly by in vitro Vero cell assay, by an immunoagglutination test kit using sensitized latex or by ELISA or indirectly by PCR techniques to detect the corresponding genes (stx1 or stx2). All these alternative tests carried out on pure cultures and hence can only be preformed after different steps including pre-enrichment, enrichment, isolation and purification. In the proposed technique, polymixin B will be added to stimulate toxin production by putative EHEC during the enrichment step to allow direct detection of the toxin(s) if any in enrichment broth.

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L5: Entry 15 of 57 File: PGPB Oct 31, 2002

DOCUMENT-IDENTIFIER: US 20020160020 A1
TITLE: Enterohemorrhagic escherichia coli vaccine

Summary of Invention Paragraph:
[0034] Thus, in one embodiment, the invention is directed to a vaccine composition comprising an enterohemorragic Escherichia coli (EHEC) cell culture supernatant and an immunological adjuvant. In certain embodiments, the EHEC is EHEC O157:H7 and/or EHEC O157:NM. In additional embodiments, the immunological adjuvant comprises an oil-in-water emulsion, such as a mineral oil and dimethyldioctadecylammonium bromide. In yet additional embodiments, the immunological adjuvant is VSA3. The VSA3 may be present at a concentration of about 20% to about 40% (v/v), such as at a concentration of 30% (v/v).

CLAIMS:

1. A vaccine composition comprising an enterohemorragic Escherichia coli (EHEC) cell culture supernatant and an immunological adjuvant.

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L2: Entry 5 of 5 File: USPT Nov 21, 1995

DOCUMENT-IDENTIFIER: US 5468639 A
TITLE: Isolated DNA molecule encoding ShET2 of Shigella flexneri 2a

Detailed Description Text (35):
The fluid to gut length recorded in the rabbit ileal loops, 0.5 ml/cm, measured using graduated syringes (fluid) and a scale (length), was substantially less than seen with enterohemorrhagic E. coli (EHEC) strain 933J, serotype (0157:H7), where ratios of 1.5-2.0 ml/cm occur. However, the recorded fluid to gut length measured using graduates syringes (fluid) and a scale (length) still represents definite evidence of net secretion and fluid accumulation. On histologic examination of the sections of intestine from each loop, severe tissue damage was observed with whole cultures of M4243, characterized by prominent necrosis of the luminal epithelium and marked villus atrophy. In contrast, with M4243 sterile culture supernatant, no tissue damage was detected. Further, no tissue damage was observed with whole cultures of M4243avir or sterile supernatants therefrom. Moreover, no tissue damage was observed with tissue incubated with the negative control strain HS.

Detailed Description Text (60):
Whole culture supernatants and cell lysates of enterohemorrhagic E. coli (EHEC) strain 933J, serotype 0157:H7, which elaborates Shiga-like toxin 1 (SLT1), were used as the positive control in the Vero cell cytotoxicity assay (Fasano et al, Infect. Immun., 58:3717-3723 (1991)). The whole supernatant of non-pathogenic E. coli strains HS, which has been used extensively as a negative control in assays of pathogenicity and in clinical studies (Levine et al, Lancet, I:1119-1122 (1978); and Levine et al, J. Infect. Dis., 148:699-709 (1983)), was used as a negative control in the Vero cell cytotoxicity assay.

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VACCINATE, DON'T PROCRASTINATE

Vaccines directed at the toxic component of bacterial pathogens have proven quite effective in preventing certain diseases. Most licensed toxoid ***vaccines*** are relatively crude, but effective, preparations. These **vaccines** consist of partially purified toxin preparations obtained from ***culture*** supernatants of bacteria such as C. diphtheriae, C. tetani, or B. anthracis. Formaldehyde treatment is used to detoxify the diphtheria and tetanus toxins for ***vaccine*** formulation. The anthrax **vaccine** contains the protective antigen and small amounts of the lethal factor and edema factor toxins. The current botulinum ***vaccine*** is an investigational drug composed of crude preparations of five botulinum toxoids and is distributed by the Centers for Disease Control and Prevention to researchers that work with the toxin or organism. Acellular pertussis **vaccines** that contain pertussis toxoid, alone or as one of several components, are as effective as killed whole-cell **vaccines** but less reactogenic (73); such **vaccines** have recently been approved for use in infants as well as older children.

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L1: Entry 25 of 54 File: PGPB Oct 31, 2002

DOCUMENT-IDENTIFIER: US 20020160020 A1
TITLE: Enterohemorrhagic escherichia coli vaccine

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0012737270 BIOSIS NO.: 200000455583
Human response to Escherichia **coli** O157:H7 infection: Antibodies to **secreted** virulence factors
AUTHOR: Li Yuling; Frey Elizabeth; Mackenzie Andrew M R; Finlay B Brett (Reprint)
AUTHOR ADDRESS: Biotechnology Laboratory, University of British Columbia, 6174 University Blvd., Room 237-Wesbrook Building, Vancouver, BC, V6T-1Z3, Canada**Canada
JOURNAL: Infection and Immunity 68 (9): p5090-5095 September, 2000
2000
MEDIUM: print
ISSN: 0019-9567
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: **Vaccination** has been proposed for the prevention of disease due to **enterohemorrhagic** Escherichia **coli** (**EHEC**), but the immune response following human infection, including the choice of potential antigens, has not been well characterized. To study this, sera were obtained from five pediatric patients with acute diarrhea caused by E. ***coli*** O157:H7 0, 8, and 60 days after hospitalization. These sera were used to examine the immune response to four different **EHEC** virulence factors: Tir (translocated intimin receptor, which is inserted into the host cell membrane), intimin (bacterial outer membrane protein which binds to Tir), EspA (**secreted** protein which forms filamentous structures on **EHEC** surface), and EspB (inserted into the host membrane and cytoplasm). The response to O157:H7 lipopolysaccharide was also examined. Sera were assayed against purified recombinant proteins using immunoblot analysis and by enzyme-linked immunosorbent assay to determine the sera's titers to each of the antigens in all patients. We found that there was little reaction to EspA, EspB, and intimin in the acute-phase sera, although there was some reactivity to Tir. By day 8, titers of antibody to all four virulence factors were present in all patients, with a very strong response against Tir (up to a titer of 1:256,000), especially in hemolytic-uremic syndrome patients, and lesser strong responses to the other three antigens. The titer to the antigens 60 days after hospitalization was decreased but was still highest for Tir. These results suggest that there is a strong immune response to Tir, and to a lesser extent to the other three virulence factors, following **EHEC** disease, indicating that these bacterial molecules are potential ***vaccine*** candidates for preventing ***EHEC*** disease. They also suggest that bacterial virulence factors that are inserted into host cells during infection by type III **secretion** systems (Tir or EspB) are still recognized by the host immune response.

L5: Entry 52 of 57 File: DWPI Mar 1, 2005

DERWENT-ACC-NO: 2002-557723
DERWENT-WEEK: 200568

TITLE: Vaccine composition useful for eliciting immunological response in ruminant and for reducing colonization or shedding of enterohemorragic Escherichia coli, comprises enterohemorragic E. coli cell culture supernatant

Basic Abstract Text (1):
NOVELTY - A vaccine composition (I) comprises an enterohemorragic Escherichia coli (EHEC) cell culture supernatant (CCS) and an immunological adjuvant.

Standard Title Terms (1):
VACCINE COMPOSITION USEFUL ELICIT IMMUNOLOGICAL RESPOND RUMINANT REDUCE COLONY SHED ESCHERICHIA COLI COMPRISE COLI CELL CULTURE SUPERNATANT

0013521797 BIOSIS NO.: 200200115308
Diarrhoeagenic Escherichia **coli**: An emerging problem?
AUTHOR: Clarke Stuart C (Reprint)
AUTHOR ADDRESS: Scottish Meningococcus and Pneumococcus Reference Laboratory, Department of Microbiology, North Glasgow University Hospital NHS Trust, Stobhill Hospital, Balornock Road, House on the Hill, Glasgow, G21 3UW, UK**UK
JOURNAL: Diagnostic Microbiology and Infectious Disease 41 (3): p93-98
November, 2001 **2001**
MEDIUM: print
ISSN: 0732-8893
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: Diarrhea remains one of the main sources of morbidity and morbidity in todays world and a large proportion is caused by diarrheagenic Escherichia ***coli*** . They are a particuar problem in developed countries although traveler's diarrhea and hemorrhagic colitis are also a problem in developed countries. There are seven classes of diarrheagenic E. ***coli*** , namely enteropathogenic E. ***coli*** (EPEC), ***enterohaemorrhagic*** E. ***coli*** (***EHEC***), enteroinvasive E. ***coli*** (EIEC), enterotoxigenic E. ***coli*** (ETEC), enteroaggregative E. ***coli*** (EAggEC), diarrhea-associated hemolytic E. ***coli*** (DHEC) and cytolethal distending toxin (CDT)-producing E. ***coli*** . Many of their virukence determinants have been determined and some classes of diarrheagenic E. ***coli*** produce toxins. The virulence factors of some diarrhogenic E. ***coli*** have yet to be full determined and in the meantime they remain a large and emerging problem without the availability of effective ***vaccines*** .

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